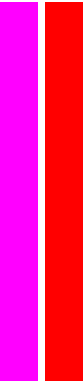


Mestrado  
Medicina Tradicional Chinesa

# Time-frame Assessment of Acupuncture Effects: How Fast Does Acupuncture Work?

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## **TIME-FRAME ASSESSMENT OF ACUPUNCTURE EFFECTS: HOW FAST DOES ACUPUNCTURE WORK?**

Dissertação de Candidatura ao grau de Mestre em Medicina Tradicional Chinesa submetida ao Instituto de Ciências Biomédicas de Abel Salazar da Universidade do Porto.

Orientador: Doutor Jorge Pereira Machado

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## Abstract

**Objective:** To assess the response time of acupuncture. To demonstrate that acupuncture has an instant effect in changing electro-dermal conductivity in specific acupoints, through a randomized controlled trial. Additionally, to indicate through a case-study that acupuncture may be an excellent choice in order to reduce the recovery time of injuries from dental medicine interventions.

**Methods:** (1) *Trial:* The experiment was designed as randomized blinded control trial, with 56 volunteers. A set of acupoints designated as Reactive Electrical Permeable Points (REPP – mostly Yuan points) was measured in terms permeability to electric current. Patients with a split-meridian (difference between left and right measurement) were randomly included in the control (no acupuncture) or experimental group (insertion of only one needle). After five minutes of needle insertion in a specific “correcting” point (Luo point), the needle was removed and REPPs were re-measured twice with a five-minute interval.

(2) *Case-Study:* A 49 years old female was submitted to a dental implantation at the location of the second mandibular premolar at the right side, with consequent permanent paresthesia in chin and lip. Distal acupuncture was used according to the balance method. The patient was submitted to acupuncture sessions, once per week, during four weeks

**Results:** (1) *Trial:* From the 56 volunteers, 32 had a valid split-meridian difference ( $SMD > 20 \mu A$ ) and were included in the study (initial measurement – M1). The results indicate that 85% of the experimental group exhibited a  $SMD < 20 \mu A$  and 45% had a  $SMD = 0 \mu A$  after the second measurement (M2 – after puncture and needle removal). At the third measurement (M3), 100% of the experimental group had  $SMD < 20 \mu A$  and 55% was with a  $SMD = 0 \mu A$ . In contrast, for the control group, 75% and 92% remained with a  $SMD > 20 \mu A$ , at M2 and M3 respectively, when compared with the initial measurement (M1). Results obtained between the experimental and control group are significantly different ( $p < 0.0001$ ).

(2) *Case-study:* After the acupuncture treatments, it was observed a full recovery in four weeks, with reduction of the area affected within the session time (45 min.). In contrast, the probable recovery time of the paresthesia, which was indicated by the doctor, was between six month and one year.

### **Conclusions:**

Through the trial, the present exploratory work indicates that there is evidence of instant body response to acupuncture that can be detected by changes in the electro-conductivity of specific acupoints. Additionally, we observed a fast response (four weeks) in chin and lip paresthesia reduction of a patient that suffered nerve trauma after dental intervention, when compared with the time response expected (six month to one year).

Keywords: Ryodoraku, Split-Meridian Difference, Yuan/Luo, Electro-conductivity

## Resumo

**Objectivo:** Avaliar o tempo de resposta de acupunctura. Demonstrar que a acupunctura tem um efeito imediato mensurável pela variação de condutividade elétrica em pontos específicos. Adicionalmente, indicar que a acupunctura poderá ser uma escolha favorável no sentido de reduzir o tempo de recuperação de lesões consequentes de intervenções de técnicas de medicina dentária.

**Métodos:** A experiência foi concebida como um estudo clínico randomizado com grupo de controlo e cego para os 56 voluntários que participaram. Um conjunto de pontos de acupunctura designados por Pontos Reativos Eletricamente Permeáveis (REPP (*Reactive Electro-Permeable Points*)) – correspondendo à maioria dos pontos Yuan) foram medido em termos de permeabilidade à corrente elétrica. Os pacientes que apresentavam valores diferentes entre os pontos medidos nos membros esquerdo e direito (*“split-meridian difference”* - SMD) foram incluídos aleatoriamente no grupo de controlo (sem acupunctura) ou no grupo experimental (intervenção com apenas uma agulha). Cinco minutos após a inserção da agulha no ponto de “correção” (ponto Luo-Conexão), a agulha foi removida e os REPPs foram reavaliados duas vezes com um intervalo de cinco minutos entre medições.

Estudo de caso: Paciente do sexo feminino de 49 anos foi submetida a um implante dentário no local do segundo pré-molar inferior do lado direito, resultando numa parestesia permanente no queixo e lábio. Foi aplicada acupunctura distal de acordo com o método do equilíbrio. A paciente foi submetida a sessões de acupunctura, uma vez por semana, durante quatro semanas.

**Resultados:** Dos 56 voluntários, 32 tinham um SMD válido ( $SMD > 20 \mu A$ ) e foram incluídos no estudo (M1 – medição do tempo inicial). Os resultados indicam que 85% dos pacientes do grupo experimental apresentaram um  $SMD < 20 \mu A$  e 45% tinha um  $SMD = 0 \mu A$  na segunda medição (M2 – após punção e remoção da agulha). Na terceira medição (M3), 100% dos pacientes do grupo experimental tiveram  $SMD < 20 \mu A$  e 55% apresentavam um  $SMD = 0 \mu A$ . Por outro lado, 75% e 92% dos pacientes do grupo de controlo permaneceram com um  $SMD > 20 \mu A$ , após a segunda (M2) e terceira medição (M3), respectivamente, quando comparado com a medida inicial (M1). A análise estatística dos resultados obtidos entre o grupo experimental e o grupo de controlo indica que são significativamente diferentes ( $p < 0.0001$ ).

Estudo de caso: O tempo de recuperação da parestesia, estimado pelo médico dentista, foi de seis meses a um ano. Com a aplicação da acupuntura observou-se uma recuperação completa em quatro semanas, em alguns casos com redução da área afetada durante a sessão de tratamento (máx. 45 min.).

### **Conclusões:**

O presente trabalho experimental indica que há evidência de uma resposta fisiológica instantânea à acupuntura, e que pode ser detectada por alterações na condutividade elétrica de pontos de acupuntura específicos. Em adição, observou-se uma resposta rápida (quatro semanas) na redução da parestesia no lábio e queixo da paciente, quando comparado com o tempo de resposta esperado (seis meses a um ano).

Palavras-chave: Ryodoraku, Diferença da Assimetria de Meridianos, Yuan/Luo, Electro-condutividade

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## Abbreviations

AIC – Akaike Information Criterion

H0 – Null hypothesis

H1 – Testing hypothesis

Mi – Measurement of the REPP in time, with  $i=1,2,3$

REPP – Reactive Electro-Permeable Points

SMD – Split-Meridian Difference

TCM – Traditional Chinese Medicine



## Preface

I will never forget the first time I have seen a graphical representation of a human being with “lines and dots” all over the body – a representation of the classical teaching of meridians and acupuncture points, which is a core subject in Traditional Chinese Medicine (TCM). This incident happened 19 years ago, at a school library. Since then, the curiosity about Chinese medicine never stopped growing and the need for learning more on the subject remains until now.

Simultaneously to the studies that led to my PhD degree in Chemical and Biological Engineering at the University of Minho, I was also studying TCM in the Portuguese Institute of Naturology (IPN - Instituto Português de Naturologia). Thus, in one hand I have developed the scientific thinking to look at things in a fully objective way, while in the other hand I was torn with the need to believe in theories not yet fully explained by scientific means, sometimes even strange – but fully supported by results obtained in clinical practice. It might have been the reason that made me search for objective methods in Chinese Medicine, but never forgetting the core teachings – actually, my personal motivation was to prove the core theories through scientific methodology.

The need to find a scientific explanation of the empiric language of the classic Chinese medicine brought me to the Master course of TCM at the Institute of Biomedical Science Abel Salazar – University of Porto. Here, a scientific view of the Chinese medicine was taught within the Heidelberg Model of TCM. Several terms of the empirical language used in TCM were fully explained under science light. As an example, “heat” and “cold” or “excess” and “deficiency” patterns in TCM were fully explained under physiological mechanisms that brought me a new vision of how to think when using the rationale of TCM core concepts.

Within the several areas of TCM (e.g. Phytotherapy, Acupuncture, Tui Na, Qi Gong), acupuncture was the one that primarily took my attention. Thus, my research theme for the present thesis is related with acupuncture and meridian research. Research in meridian theory has been very controversial, and it still needs a lot of progressive work. That is why I am orienting my efforts in scientific work that call help to prove the existence and/or physiological mechanisms of these “theoretical paths”, which actually may really exist physically.



Thus, the aim of this thesis is first to demonstrate that the body must have short-term (e.g. minutes) response to needle puncture that should be detected through electrical conductivity change in specific acupoints called *Yuan* (source) points. The physiological mechanism and possible relation to “physical meridians existence” is still under research, and thus not explained here.

The central work of the present thesis is fully based on the work published in the *World Journal of Traditional Chinese Medicine*, entitled: “**Assessment of short-term acupuncture effect through electro-conductivity variation of Yuan-Source acupoints**”. This work was also presented in one oral communication at the World Congress of Chinese Medicine 2015 in Barcelona - Spain, under the same title and in a panel communication in the 1st Luso-Chinese Forum for The Development of Chinese Medicine in Cascais, Portugal under the title: “Short-term acupuncture effect assessed by changes in acupoints electrical conductivity: a blinded control trial”. Additionally, and with the objective of showing that acupuncture may induce faster results than the actual western state of the art in certain injuries, a case study has been added.

I hope that the lecture will be pleasant and above all that it may add one step further in the understanding of features of the acupuncture technique.

I would like to thank several persons that contributed the present work. I will start by my thesis supervisor, Professor Jorge Machado that was always available to discuss all the work produced and pushed forward to develop a good quality research. I also thank all my colleagues that participated in the articles that resulted from the present research work, namely, André Santos, Rui Pedro Loureiro and Rui Gonçalves. I also thank professor António Cabrita for his valuable contribution in the case study publication. I thank my beloved wife for all her effort in supporting me in this process.

## **Chapter 1.**

### **Motivation and Objectives**



The practice of Traditional Chinese Medicine that includes acupuncture, phytotherapy and “tui na” (massage) is a growing practice in western countries<sup>1</sup>. However, there is a need to look at the millenary and empiric language used within Chinese philosophy and to validate the evidences with a scientific approach. For years, due to the lack of legislation in Portugal, practitioners with no scientific knowledge or background started to use acupuncture or other TCM techniques even with few lectures on the subjects. Thus, there is a need to change the way TCM is being practice clinically to guarantee the safety and wellbeing of the patients. Additionally, and to allow an integrative medicine approach, practitioners of TCM should develop a curriculum in both western life science and TCM philosophy. Thus, the teaching of TCM must include western life science classes as well as well as research and development applied to TCM.

As TCM rational is fully based in observation and evidence, it should be possible to describe it in a scientific way of thinking, within the knowledge developed in western medicine and life science. In oriental countries, hundreds of articles are published annually in scientific journals. However, there is still an emerging need of scientific research in TCM in western countries. The Institute of Biomedical Sciences Abel Salazar – University of Porto, is at the moment the sole institution approved by the national education ministry with a master course in TCM within the Heidelberg model. Thus, it is also the sole institution in Portugal delivering scientific research whether through master thesis or through published articles in peer-reviewed journals.

Evidence-based research can easily be applied to several therapeutic techniques used in TCM. Specifically in acupuncture, hundreds of articles are being publish with researchers from western countries. For example, PubMed results for the word “acupuncture” is 25218 entries<sup>2</sup>. Specifically in pain management, the search for: “acupuncture” AND “pain”, also in PubMed, resulted in 6179 entries<sup>2</sup>.

The majority of published trials using acupuncture for pain reduction usually use local acupuncture as method. Additionally, as a variation of the needling technique, electro-acupuncture or laser-acupuncture may be used<sup>3,4</sup>. In these cases, pain reduction happens during several sessions and with several days of treatments to achieve a relevant pain reduction. However short-term results obtained through acupuncture in pain management have being described in books and research articles – mainly using the balance method reasoning<sup>5-7</sup>. In clinical context, we have observed hundreds of cases of pain reduction after

acupuncture application that occurred in a time frame of few minutes (one to five minutes), several times to a full reduction of pain sensation. In some clinical cases, the pain reduction happened in less than a minute, appearing to have an “instant result”. In all these cases, where acupuncture achieve a fast pain reduction, the needling was made in distal points in relation to the pain location, and thus, never in the pain area.

These clinical observations (“instant pain reduction”) compared with the published research using local acupuncture for pain reduction made us understand that the response time of acupuncture is different depending on the location of the needle in reference to the pain (local puncture vs distal puncture). The reasoning on the time that acupuncture take to have a measurable effect (in this case in pain scale) was the starting point to the present work.

From that point of view, several questions were made:

- In how much time does acupuncture delivers a measurable effect?
- Does acupuncture have an instant effect in a certain part of the body that is specific to the acupoints used (ex: local vs distal acupuncture for pain reduction)?
- One group of acupoints may have a faster result compared to a different group of acupoints?
- How can we measure the response time of acupuncture beside the use of subjective methods (ex: pain scale measurement)?
- Can acupuncture have faster results than the state of the art in injury management (whether the result is pain or altered sensation due to nerve injury)?

The first challenge was to find a measurable non-subjective variable that could change in time due to acupuncture application. Scales are used to measure pain intensity, however it is still a very subjective way of measuring<sup>8</sup>. Thus, a different way of measuring acupuncture's effects that would not be dependent on the patient's own perception were explored. Several studies using MRI have already been developed to show the effect that needling has in central nervous system<sup>10-12</sup>. However, TCM research in Portugal has still a low budget and at the moment, to have access to MRI technology for research in TCM is highly difficult. Thus, less expensive techniques were examined to evaluate the potential action of acupuncture. We have found that electro-dermal conductivity measurement related to TCM has been used as diagnosis and was called as Ryodoraku (meaning “line of high conductivity”), by a Japanese nephrologist in the 50's – Dr. Nakatani<sup>13</sup>. He has found that patients with nephritis would exhibit a high electro-dermal conductivity in points located in the medial side of the ankle and along the medial side of the leg. The high conductive points that Dr. Nakatani

found were very close to the location of the classical trajectory and point known as the TCM kidney channel (or meridian). Later he has described other trajectories very similar to the known TCM channels after finding several points with different electro-dermal conductivity along the whole body<sup>13</sup>. In classic TCM, there are 14 main superficial channels described with a specific number of acupoints. Dr. Nakatani's work described those points very closely. Additionally, he indicated that specific values would be correlated to certain diseases. From this knowledge, we have decided that this type of technology could be useful to measure changes in electro-dermal conductivity as an effect of acupuncture practice. The next challenge was to decide which point would be measured and which would be punctured as cause-effect scenario. Following the work of Dr. Nakatani, he described that the specific category of acupoints, called Yuan (source), that would reflect the mean values of electrical skin conductivity, and that these point would be representative of the whole channel. Thus, Yuan acupoints were selected as the measuring points (additional information on the location and importance of these points is given in chapter 3 – Table 3.1). The puncturing points selected were the category points called Luo (connecting), which are distal points in relation to the Yuan points, and are described as balancing points of the channels. All the details on these points are explained in Chapter 3. At this point, the method for measuring the effect of acupuncture in time was defined. A controlled blinded randomized trial was defined and executed. The methods, results and discussion are fully presented in Chapter 3, as well as published in an international peer-reviewed open-access journal<sup>14</sup>, as indicated in Chapter 2.

Thus, the main objective of the present work was to measure the response time of acupuncture – that is, to measure an effect through a non-subjective variable (electro-dermal conductivity) caused by acupuncture practice. Under the rationale of testing the response time of acupuncture effects, a specific objective was to compare through a clinical case study if acupuncture can achieve faster results than the actual state of the art western procedures, in the specific case of altered sensation due to nerve injury. In this study, we indicate acupuncture has a potential therapy to accelerate the recovery process of a person with chin and lip paresthesia that was injured during a dental procedure, when compared with actual procedures to achieve the same results. A case study is fully described in Chapter 4 of the present thesis and also published as short-communication in an international peer-reviewed journal<sup>15</sup> as indicated in Chapter 2 that addresses the use of acupuncture to accelerate the patient's recovering of sensations previously lost by mandibular nerve injury in a dental implantation procedure.

The legislation in Portugal is changing and TCM practitioners have now the legal right of practicing acupuncture and other TCM therapies with deontological independence<sup>16-17</sup>. This also requires better-trained practitioners with a higher background in science production in TCM field. Thus, we additionally expect that the present work in the thesis form of in the form of the already published articles can contribute in the growing science developed in ICBAS-UP in TCM field. We additionally expect that the present research in acupuncture's effects as function of time can stimulate future students to develop high-quality research in this research branch of TCM.

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## Chapter 2.

### Thesis Overview



After defining the motivation and objectives of this project in **Chapter 1**, the internal structure of the thesis is presented as follow:

**Chapter 3** addresses an acupuncture based controlled trial with 53 participants. The objective of the trial was to understand if acupuncture effect could be measured through changes in electro-conductivity variation of specific acupoints, in a short time frame (5 to 15 minutes). The results show a statistically relevant correlation.

**Chapter 4** addresses the results obtained with acupuncture in chin and lip paresthesia elimination in a very short time frame when compared with conventional treatments. Thus, it points to the possibility that acupuncture can induce fast recovery of altered sensations due to nerve injury after a dental procedure. It also indicates the potential application of acupuncture in dental medicine.

**Chapter 5** is the concluding chapter and it sets several questions that are important to address in future work, following the present one.

The work produced during the master course resulted in two published articles, one oral communication and one panel communication (poster), as indicated below:

**Articles:**

1. Ribeiro DC, Santos A, Loureiro RP, Gonçalves R, Machado J. 2015. Assessment of short-term acupuncture effect through electro-conductivity variation of Yuan-Source acupoints. *World J. Tradit. Chin. Med.* 1(4): 48-52. DOI: 10.15806/j.issn.2311-8571.2015.0023.
2. Ribeiro DC, Cabrita AMS, Machado J. 2016. Acupuncture for paresthesia/anesthesia elimination after dental implantation complications: a case report. *Experimental Pathology and Health Science* 8(2):111-114.

**Oral communication:**

Ribeiro DC, Santos A, Loureiro RP, Gonçalves R, Machado J. Assessment of short-term acupuncture effect through electro-conductivity variation of Yuan-Source acupoints. 12th World Congress of Chinese Medicine, Barcelona, Spain, 24-26 September 2015

### **Panel Communication:**

Ribeiro DC, Santos A, Loureiro R, Gonçalves R, Machado J. Short-term acupuncture effect assessed by changes in acupoints electrical conductivity: a blinded control trial. 1st Luso-Chinese Forum for The Development of Chinese Medicine, Cascais, Portugal, 27-29 September 2015.

## **Chapter 3.**

**Assessment of short-term acupuncture effects through electro-dermal conductivity variation.**



## Introduction

Meridian and acupoint theory is a central subject in Traditional Chinese Medicine (TCM) upon which acupuncture intervention is fully based. In the attempt to show that the acupuncture mechanism of action goes beyond the nervous system, research has progressed to demonstrate the existence and nature of these channels. In 1963, in North Korea, Bong-Han Kim announced a structure that could be interpreted as the commonly known meridians, and called them Bong-Han ducts<sup>1</sup>. Decades later, successful scientific works demonstrated the existence of the channels<sup>2</sup> and several properties of these channels including electromagnetic and optical properties (biophoton)<sup>3,4</sup>.

Other researchers found that several skin disorders would align in a path along the body that could be consistent with the trajectory of the meridians described in TCM<sup>5</sup>. Moreover, other researchers suggested that meridians could be related with the interstitial fluids that, in a specific channel would have a low hydraulic resistance, and that would be a vehicle of communication<sup>6</sup>. Intriguingly, it was found that an induced blockage of the low hydraulic channel corresponding to the TCM stomach meridian in a pig would induce gastrointestinal pathological changes (stomach and intestinal distention)<sup>6</sup>.

Almost one decade before the work of Bong-Han Kim, Nakatani found that patients with nephritis would exhibit an exaggerated electrical conductivity in certain points on the leg when compared to adjacent areas<sup>7</sup>. Curiously, the alignment of those permeable points to electrical current would trace a path similar to the TCM kidney meridian. Dr. Nakatani called that path ryodoraku, that means “line of high electrical conductivity”<sup>7</sup>.

Thus, past and recent studies indicate that the meridian network can conduct electricity, light and is sensible to physical pressure. Even more recently, a theoretical and exploratory work came forward with the hypothesis that meridians may have superconductor’s properties<sup>8</sup>.

An interesting information that the ryodoraku measurement can provide is the perception that a meridian can have different electrical conductivity on the left and right side, for the same acupoint. This condition is defined as a split-meridian. Despite the physiological meaning of a split-meridian is not fully understood, these responses are often found to correlate with states of disease and health<sup>9,10</sup>. The aim of the present work is to show that a split-meridian condition can be changed in a short-term period after an acupuncture procedure.



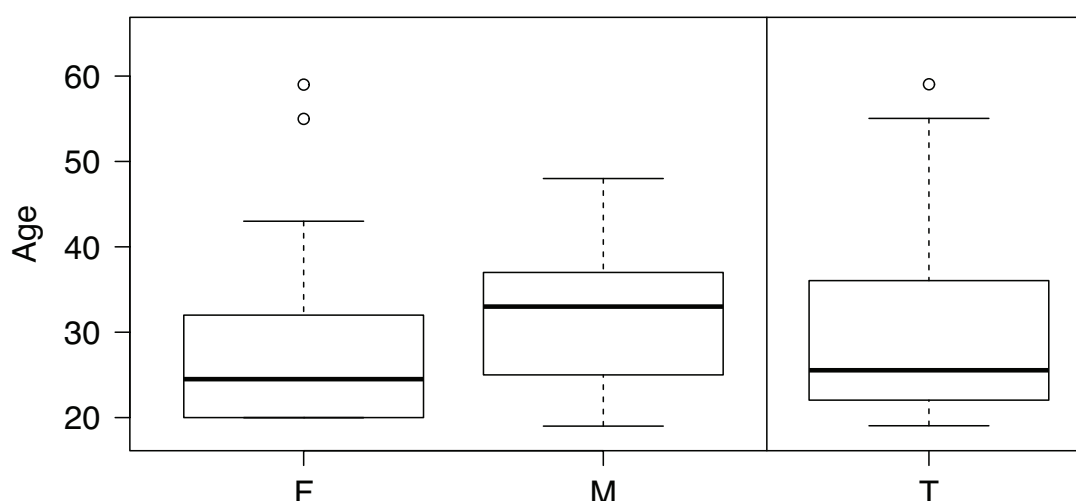
### Materials and Methods

#### *Ethics Statement*

The work described in this article was carried out in accordance with the Code of Ethics of the World Medical Association (Declaration of Helsinki) for experiments involving humans. Subjects provided written informed consent before participation in the study.

#### *Participants*

Fifty-six patients were recruited for the study: 20 were in the experimental group (16 females and 4 males), 12 were in the control group (6 females and 6 males) and 24 were excluded (see exclusion criteria). Volunteer students and workers from Institute of Biomedical Sciences Abel Salazar - University of Porto (Portugal) and Portuguese Institute of Naturology (Portugal) formed both experimental group and control group with ages between 19 and 59 years old. The statistics on age distribution for female and male participants is depicted in Figure 3.1.



**Figure 3.1 – Trial participants age distribution:** Age distribution of volunteer participants (F – female, M – Male, T – Total participants) in the present study. The minimum and maximum age all of the participants were 19 yrs to 59 yrs., being the 1st quartile: 22.5 yrs, median: 25.5 yrs and 3rd quartile: 36 yrs.

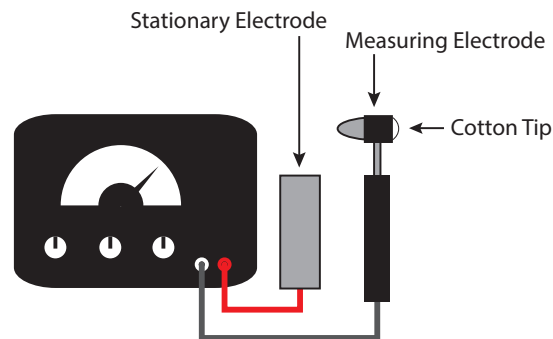
#### *Exclusion Criteria*

Patients with a non-valid split-meridian (difference between measurements in the left and right limb for the same acupoint is under 20  $\mu$ A) and/or under psychotropic medication were excluded.

### Research procedure

#### Point measurement, split-meridian definition and acupuncture procedure

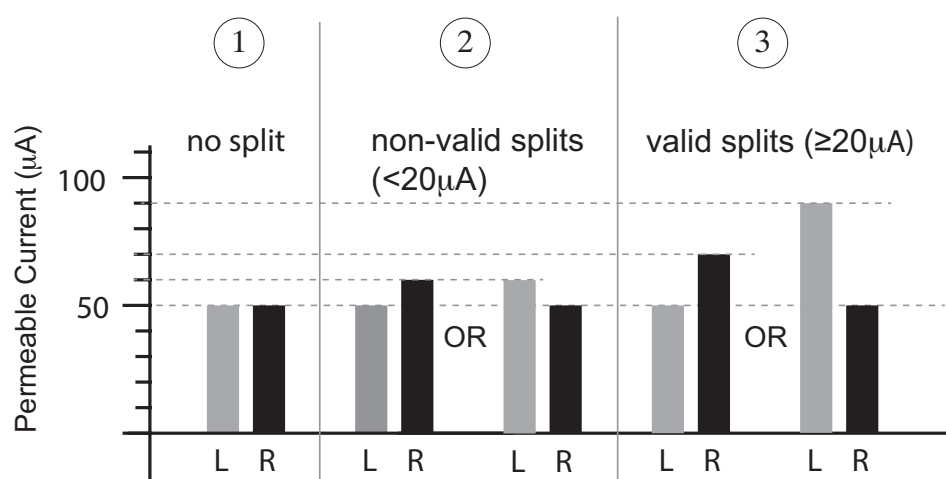
The measurement of REPP was performed with the IW-ZEN Ryodoraku neurometer. For each measurement, a 12 V direct current (DC) was used calibrated to a maximum of 200  $\mu\text{A}$ . The stationary (grip) electrode is made of stainless steel and was held in the left hand of each patient while the measuring electrode is in contact with the acupoint for a maximum time of 5 seconds per point. The measuring electrode is equipped with a conical tip with cotton embedded in a saline solution (0.9% m/m of NaCl) and it is applied in contact with the skin at the acupoint location. Figure 3.2 is a representation of the IW-ZEN Ryodoraku apparatus.



**Figure 3.2 – Schematic presentation of equipment:** Ryodoraku measuring equipment representation. Stationary electrode is held in left hand of patients while the measuring electrode contacts the REPP with the cotton tip embedded in saline solution.

The measured REPP are mainly Yuan-Source acupoints with the exception of Hégū (LI4) that was substituted by Yángxī (LI5). The twelve main channels were measured in terms of electrical conductivity (Table 3.1) and valid split-meridian was searched. A valid split-meridian was defined as when, for a specific channel, the REPP measurement of the left side was different relative to the right side by at least 20  $\mu\text{A}$ . As an example, for the Lung channel, if the measured value for LU9 (Taíyuān) at the left side is 50  $\mu\text{A}$  and the measured value for the same point at the right side is 70  $\mu\text{A}$  a valid split meridian is present.

Thus, for each measurement one can have 3 possible scenarios (Figure 3.3): 1- there is no split-meridian (left and right side measurement are equal), 2 – there is a split-meridian but it is non-valid (the difference between the left and right measurement is under 20  $\mu\text{A}$ ), 3 – there is a valid split-meridian (the difference between the left and right measurement is equal or higher than 20  $\mu\text{A}$ ). For all the 12 pairs (left and right) of acupoints (24 measurements), patients can have more than one valid split-meridian. In that case, the split-meridian with the largest difference between sides was selected.



**Figure 3.3 – Possible scenarios during measurements:** Graphical explanation of the three possible scenarios during the measurement of electrical conductivity in paired acupoints (the same acupoint in the left and right limb). Scenario 1 is when the measurement of the electrical conductivity in an acupoint in the left (L) limb is the same of the right (R) limb. Scenario 2 is when the electrical conductivity between left and right acupoints is different but under 20 µA, and is designated as non-valid split. Scenario 3 is when the electrical conductivity between left and right acupoints is different and equal or higher than 20 µA, and is designated as a valid split.

The experimental group was submitted to acupuncture in only one acupoint. The acupoint selected was always the Luo-Connecting point (Table 3.1) on the channel where the split was detected, and on the side where the lower value of electric conductivity occurs.

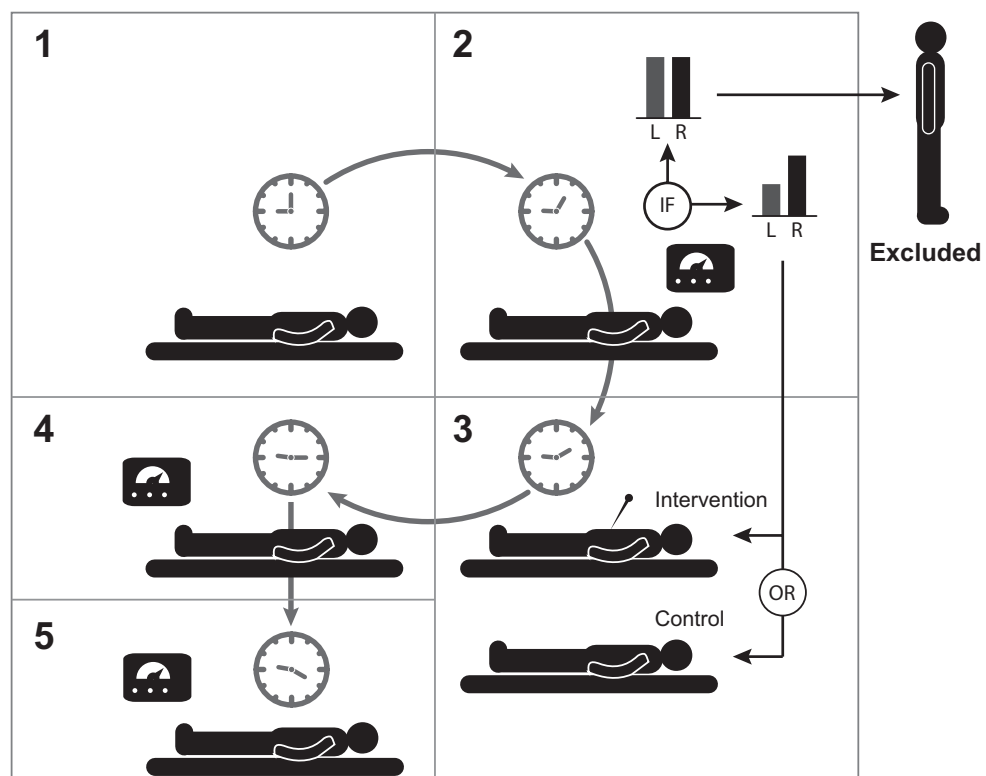
**Table 3.1 – Measured REPP for all channels and corresponding punctured Luo points (experimental group only) if valid split-meridian was detected.**

Channels (meridians)	Measuring Points (REPP)		Luo Points	
	Ref.	Name	Ref.	Name
Lung	LU9	Taiyuan	LU7	Lieque
Pericardium	PC7	Daling	PC6	Neiguan
Heart	HE7	Shenmen	HE5	Tongli
Small Intestine	SI4	Wangu	SI7	Zhizheng
Sanjiao	SJ4	Yangchi	SJ5	Waiguan
Large Intestine	LI5	Yángxi	LI6	Pianli
Spleen	SP3	Taibai	SP4	Gongsun
Liver	LV3	Taichong	LV5	Ligou
Kidney	KI3	Taixi	KI4	Dazhong
Bladder	BL64	Qiuxu	BL58	Feiyang
Gall Blader	GB40	Jinggu	GB37	Guangming
Stomach	ST42	Chongyang	ST40	Fenglong

Thus, for the example given previously (split-meridian detected in Lung channel), the electric current at LU9 - left side was 50  $\mu$ A opposed to 70  $\mu$ A at the right side. In this case, the Luo point of Lung channel (LU7 - Lìequē) was punctured at the left side (lower measured value).

### Experimental phases

The experiment was divided in five consecutive phases (Figure 3.4). In phase 1, patient was at rest in supine position for 5 minutes. In phase 2, measurements of REPP were made for the 12 channels. If at least one valid split-meridian was found then the patient was allocated to the control group or experimental group. If no valid split-meridian was found the patient was excluded from the study.



**Figure 3.4 – Trial phases exemplification:** Depiction of the experimental phases. Phase 1: Rest phase – patients rest for 5 minutes in supine position; Phase 2: REPP measurement 1 – measurement of REPP and examination of split-meridians; Phase 3 – Intervention phase: control group stay at rest for 5 minutes while experimental group is punctured for 5 minutes; Phase 4: REPP measurement 2 – measurement of REPP in both control and experimental groups and additional 5 minutes resting period; Phase 5: REPP measurement 3 – measurement of REPP in both control and experimental groups and end of experiment.

In phase 3, the patients of the control group were at rest for 5 minutes. The patients of the experimental group were punctured in the Luo-Connecting point that belongs to the channel where the split-meridian was found, and at the limb side with the lowest electrical conductivity. The needle remained inserted for five minutes and then was removed. In phase

4, both patients in the control or experimental groups were measured in the same way as in phase 2, and they stayed an additional five minutes at rest. In phase 5, both patients in the control or experimental groups were measured in the same way as in phase 2. After this measurement the experiment was ended.

### *Hypotheses and statistical tests*

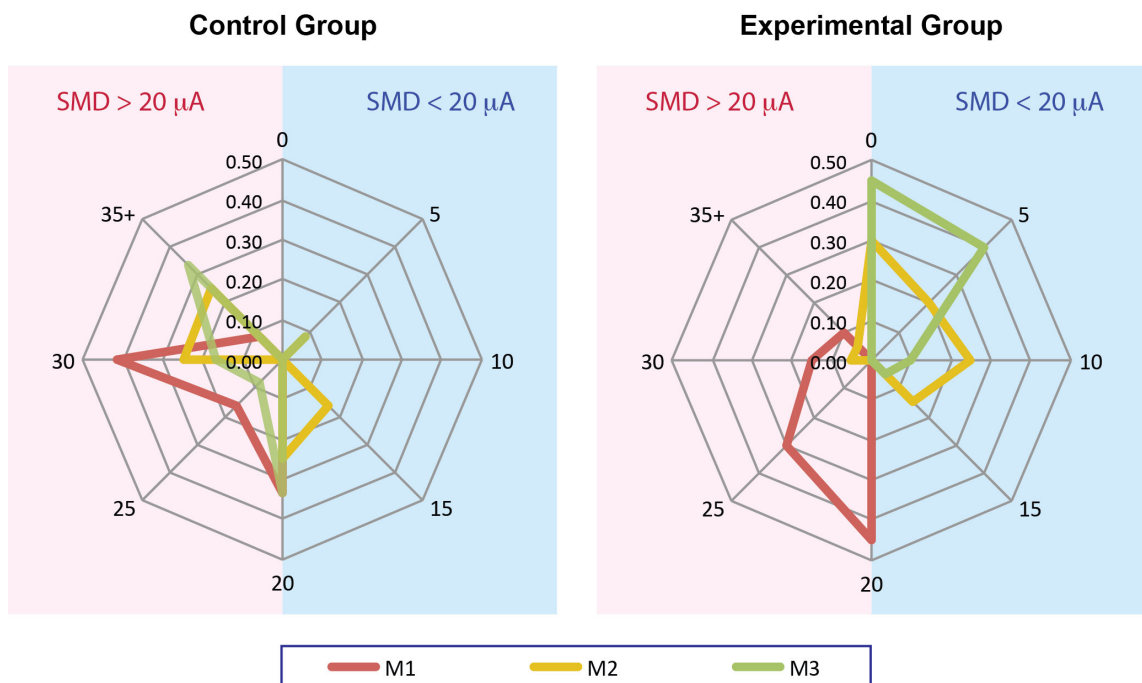
The present work aims to test the hypothesis (H1): “Patients with a split-meridian can level the values of electrical conductivity of the left and right side REPP in relative short-time period after acupuncture procedure on Luo-Connecting points”, against the null hypothesis (H0) that is: “Patients with a split-meridian will experiment no reduction of the difference in electrical conductivity between the left and right side REPP, after acupuncture, in at least the same time period tested in H1”.

The models and statistical tests used in the present work to assess the hypotheses were the linear fixed model and analysis of deviance. The dependent variable was SMD, the fixed factor was the groups and the covariate was the time. The interaction between time and group was significant due to similar values at time zero for both levels of group (experimental vs control). Individual was used as a random factor. An equivalent model without random factor was fitted and compared with the previous one by means of AIC (Akaike Information Criterion), resulting in the selection of the model with random factor (AIC=755 (random factor model) vs AIC=765) as the best model.

In order to accomplish the assumption of residual normality of the model, three outliers were removed based on the Bonferroni test for outliers. Normality was tested with Shapiro-Wilks test. The mathematical software R (v.3.1.1) was used for all the models and statistical tests.

## Results

From the 56 volunteers, 32 had a valid split-meridian and were included in the study. For the experimental group, at the moment of the first measurement (M1 – Figure 3.5, right graph) 45% of the group had a split-meridian difference (SMD) of 20  $\mu$ A, 30% of the group had a SMD = 25  $\mu$ A, 15% of the group had a SMD=30  $\mu$ A and 10% had a SMD  $\geq$  35  $\mu$ A. After the acupuncture intervention, the second measurement (M2) was performed and the results indicate that 90% of the group presented a SMD<20  $\mu$ A and 30% presented no SMD (SMD=0  $\mu$ A). The third measurement (M3 - five minutes after M2) revealed that 100% of the group had a SMD<20  $\mu$ A, 45% had SMD=0  $\mu$ A and 85% was with a split-meridian difference equal or less than 5  $\mu$ A (Figure 3.5, right graph).



**Figure 3.5 – Control and Experimental group results:** Representation of the normalized frequencies of the control and experimental groups, concerning the split-meridian differences ( $0 < \text{SMD} < 35+ \mu\text{A}$ ) for the three measurement moments of the experiment (M1 – phase 2, M2 – phase 4 and M3 – phase 5).

For the control group, at the moment of the first measurement (M1 – Figure 5, left graph) 33% of the group had a SMD of 20  $\mu$ A, 17% of the group had a difference of 25  $\mu$ A, 42% of the group had a difference of 30  $\mu$ A and 8% had a difference of more than 35  $\mu$ A. After the second measurement (M2) the results indicate that 75% of the group remained with a split-meridian with a difference equal or higher than 20  $\mu$ A. After the third measurement (M3), 92% of the group had a split-meridian difference equal or higher than 20  $\mu$ A. In all the

experiment, no patient of the control group exhibited a total reduction of the SMD ( $SMD > 0 \mu A$ ). The analysis of the linear mixed model resulted in rejection of the  $H_0$  for the factor group (Table 3.2).

**Table 3.2** - Results of the analysis of deviance performed with the linear mixed model fitted to SMD data. DF=Degrees of freedom.

Variable	DF	Chi-square	p-value
Group	1	57.2	<0.001
Time	1	41.5	<0.001
Residuals	26	-	-

## Discussion

The core interpretation of the results delivered through the present study is that Luo-Connecting acupoints have the ability to reduce the SMD value, or even eliminating the split-meridian ( $SMD = 0 \mu A$ ), in a short-time period ( $\leq 10$  minutes after needle removal). As shown in Figure 3.5, after 5 minutes of acupuncture intervention in the Luo-Connecting points, 30% of the experimental group had an  $SMD = 0 \mu A$ , which means that the split-meridian was fully eliminated. Additionally, after 10 minutes, 45% of the experimental group had no split-meridian, 85% had a  $SMD \leq 5 \mu A$  and 100% with  $SMD \leq 15 \mu A$ . Thus, the tested hypothesis  $H_1$  against  $H_0$  is significant (see Table 3.2) demonstrating that a SMD can be changed through acupuncture in corresponding Luo-Connecting point. The analysis of deviance also indicate that time is significant in the change of SMD for the experimental group (see Table 3.2).

Two important conclusions from this result are, (1) that a five minutes acupuncture intervention is enough to produce measurable changes in electrical conductivity of the skin in REPP points and (2) that the effect of reduction of the SMD is propagated in time (at least for 10 minutes – time defined in the present experimental work). The conclusion that acupuncture can have a fast physiological effect is corroborated by scientific work that shows the instant brain response to acupuncture<sup>11</sup>. As an example, researchers have found that acupuncture at GB34 (Yanglingquan) can have an instant effect in improving motor-cognition connectivity in hemiplegic stroke patients<sup>12</sup>. Thus, it is certain that responses through nervous system will happen to some extent, sometimes also including brain regions like the cerebellum and limbic-related regions<sup>13</sup>. However, several studies indicated that the brain response that exists during the acupuncture stimulation may change over time<sup>14–16</sup>. In fact, with the present work we also concluded that the acupuncture effect is propagated in time, as changes in electrical-conductivity continue to happen after needle removal. A possible

explanation for this result is that biochemical changes in the composition of the fluid in the channel (whether a Bong-Han duct<sup>1</sup> or low hydraulic resistance channel<sup>6</sup>) may happen. Changes in the composition and/or crystalline structure organization of proteoglycans associated with water molecules, specifically glucosaminoglycans (e.g. hyaluronic acid) would influence the electrical conductivity of the channel as proposed in previous work<sup>8</sup>.

Another explanation may be associated with collagen fibers. It has been proposed that the acupuncture system may be related to the collagen liquid crystalline continuum in the connective tissues that can perform a fast semi-conduction of protons through a layer of structured water molecules<sup>17</sup>. However, these results must be interpreted carefully until *in vivo* analysis of the composition of these channels is preformed paired with electrical conductivity measurements.

From the present work several questions arise such as: does the nervous system send information for the local channel cells to change the biochemical composition of the interstitial fluid? Or does it happen by information propagated through the channel itself? Or is this a mixed response? Additionally, what are the implications of a re-established electrical conductivity to pathological conditions? Are these short-term changes ubiquitous in acupuncture points or is this a phenomenon particular to the Yuan-Luo relation? What other variables may affect both the extent and the speed of the observed corrections? Further work will be oriented to give insight on these questions.

### Conclusions

From the present work we can conclude that the Luo-Conneting points have the ability to reduce or even eliminate the SMD of their corresponding channel in a short-time period (5 to 10 minutes). Additionally, after needle removal, the effect of SMD reduction is propagated in time. However, further work is needed to understand the physiological mechanisms of action of these points and implications in human health.



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## **Chapter 4.**

### **Shortening the recovery time of lesions in dental medicine – A case report.**



## Introduction

Mandibular nerve trauma due to dental procedures can result in temporary or permanent anesthesia (absence of all sensory modalities) or paresthesia (an abnormal sensation)<sup>1</sup>. These altered sensations after dental interventions (e.g. teeth removal, implants, surgery) have been extensively studied and reported<sup>2,3</sup>. Quality of life of patients that undergo such altered sensations is highly reduced. Conventional treatment may include surgical or non-surgical treatments<sup>4</sup>, use of medication (steroids)<sup>5</sup> and acupuncture<sup>4</sup>. The present article reports a clinical case of a patient that experienced anesthesia of chin and lower lip (right side) after dental implantation that was treated by acupuncture. The acupuncture treatment was based on the balanced method that relies entirely on Traditional Chinese Medicine (TCM) philosophy. Although the balance method is largely used to treat pain<sup>7,8</sup>, the same approach was performed to treat anesthesia/paresthesia. The patient reported a full recovery in four-week treatment protocol (once per week). Thus this article aims to define an acupuncture protocol to treat anesthesia or paresthesia sensation on chin and lower lip due to mandibular nerve trauma.

## Methods

### Case Description

A 49 years old female was submitted to a dental implantation at the location of the second mandibular premolar at the right side. Chemical anesthesia was used for the procedure. No complications were reported during the procedure. For the following three weeks after the implantation, the anesthesia sensation in chin and lip (only right side) remained with pain irradiating to the face (posterior area). The patient asked the dental practitioner to remove the implant but the anesthesia sensation continued for several weeks at the chin and lower lip, right side. Two month after the first intervention, acupuncture procedure was initiated in 45 minutes sessions, once per week, during four weeks. The acupuncture points were selected according to the balance method reasoning.

### *Acupuncture protocol*

Under the TCM description of trajectory of the channels (meridians), the affected area (face, chin and lip) corresponds mostly to the trajectory of the Stomach and Large Intestinal channel. The balance method is fully based in TCM philosophy<sup>6-8</sup>. Under this method's reasoning, the affected channels should never be punctured locally<sup>9</sup>. Instead, the affected channels should be treated using other channels that have the ability to balance them<sup>6,7</sup>. The balance method uses several perceptive of the duality of the Yin-Yang philosophy to generate five different balancing systems. In the present clinical case we have used the system I to balance the channels that were proximal to the affected area. According to the system I, the Stomach channel is balanced by the Large Intestinal channel and vice-versa<sup>7,10</sup>. Additionally, the balance method states that points in the hands or feet can affect areas of the head<sup>7,11</sup>. Thus, we have looked for sensitive points in the hand and foot that were contra-lateral to the affected area. Thus, the points Hégǔ (LI4), Sānjiān (LI3), Èrjiān (LI2), Shāngyáng (LI1) were used in the left hand only to balance the Stomach channel of the right face (unilateral puncture). Additionally, the points Chōngyáng (ST42), Xiàngǔ (ST43) and Nèitíng (ST44) were used to balance the Large Intestinal. For the third and fourth treatment, we added additional points of the Liver channel located in the right foot. This choice was made has the remaining anesthesia was more located on the lip, which is more related with large intestinal channel, and that according to the balance method's system II, can be treated also with Liver channel points. Points like Dàdūn (LV1), Xíngjiān (LV2), Tàichōng (LV3) were punctured homo-laterally according to the balance method<sup>7</sup> and thus additionally added to the protocol. Table 1 indicates the points used in the present protocol in detail.

**Table 4.1 – Acupuncture points used to treat anesthesia/paresthesia sensation in chin and lower lip in the present study case.**

Acupuncture Point		Puncture side*
Name	Reference	
Shāngyáng	LI1	Contralateral
Èrjiān	LI2	Contralateral
Sānjiān	LI3	Contralateral
Hégǔ	LI4	Contralateral
Chōngyáng	ST42	Contralateral
Xiàngǔ	ST43	Contralateral
Nèitíng	ST44	Contralateral
Dàdūn	LV1	Homolateral
Xíngjiān	LV2	Homolateral
Tàichōng	LV3	Homolateral

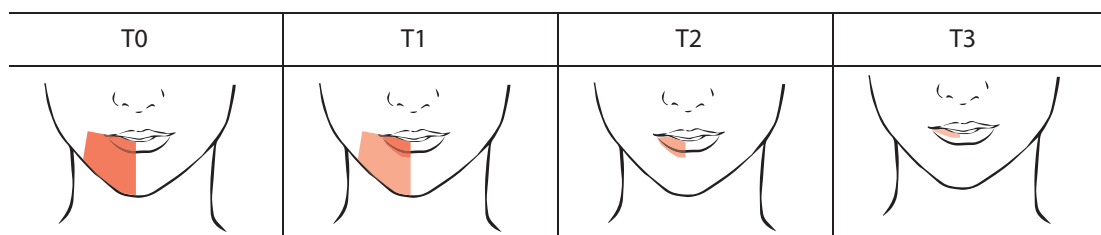
\* Puncture side relatively to the altered sensation location

The duration of each acupuncture treatment was 45 minutes and was repeated in four consecutive weeks, once per week.

## Results

A graphical explanation of the change in sensation with time (T0 to T3) is depicted in Figure 1, according to the patient's description. After the first treatment the patient referred a small increase in sensation in the chin (Figure 4.1 – T1), but also reported that the anesthesia feeling at the lip was unchanged. During the second treatment a full recovery of the sensation at the chin was felt immediately after the session (T2). Additionally, the lip sensation changed from anesthesia to paresthesia indicating some recovery of the sensation in the area. At the third treatment, the patient referred a gradual elimination of the paresthesia sensation at the lower lip and only a residual paresthesia sensation remained at the upper part of the lower lip (T3). After the fourth session, seven days after the third treatment, the patient referred a full elimination of the remaining paresthesia sensation in the lip (T4 not shown).





**Figure 4.1 – Anesthesia sensation evolution in time (T0 to T3) described by the patient.** Previous to any treatment (T0) the anesthesia sensation was located in right side of the chin and lower lip as described. Seven days after the first treatment, the anesthesia sensation in the chin has changed but remained unchanged in the lip (T1). At the end of the second treatment the anesthesia sensation of the chin was eliminated while in the lip some paresthesia sensation remained (T2). Seven days after the second treatment the patient reported a gradual elimination of the paresthesia sensation at the lower lip and only a residual paresthesia sensation remained (T3). Seven days after the last treatment, the patient reported a full recovery with no paresthesia sensation (T4 – not depicted).

Thus, after the four treatments a full recovery of the sensation in both chin and lips were reported. Six month after the treatment no regression of the state of sensation was reported. The patient indicated a full recovery of quality of life in relation to the anesthesia/paresthesia feelings.

## Discussion

The understanding of the mechanisms of actions of acupuncture treatments is still in progress<sup>12</sup>. It has been proved that acupuncture can reduce or even eliminate pain. Some acupuncture procedures using electro-acupuncture induce an anesthesia state and thus a consequent pain reduction<sup>13</sup>. The mechanism of action of electro-acupuncture has been extensively explained elsewhere<sup>14–16</sup>. However, in the present case report, we present an elimination of a state of anesthesia due to mandibular nerve trauma. The aim of the presentation of this study case is to define a possible protocol of treatment of anesthesia or paresthesia of patients that suffered a mandibular nerve trauma due to dental procedure intervention, regardless of the mechanism of action. The case study indicates a gradual improvement between treatments. Although this gradual improvement of the altered sensation may occur naturally, we also reported a sudden change in paresthesia feeling in one specific treatment (Figure 4.1 – T2) in a time window of 30 minutes. Natural changes in paresthesia are expected to occur in a superior time window (weeks or months) or may not occur at all, resulting in a permanent anesthesia or paresthesia sensation.

The balance method is known to induce a fast pain reduction (minutes to one hour) with no use of electro-acupuncture and never needling the affected area. This could explain the fast reduction of paresthesia felt in T2. This case report sets the basis for future research in the validity of this specific acupuncture treatment protocol for reduction or elimination of anesthesia/paresthesia due to mandibular nerve trauma. Additionally, it may stimulate the research in understanding the mechanisms of action of distal acupuncture points to treat physical disorders.

### Conclusions

A 49 years old woman had a mandibular nerve trauma with consequent anesthesia felling in half chin (right side) and lower lip (right side). A full recovery of anesthesia and paresthesia sensation was achieved within four weeks of acupuncture treatment. Evidence that the reported recovery was achieved due to acupuncture treatment is based on a fast reduction in paresthesia (in 30 minutes) at the second session of acupuncture. However, further studies are needed to explain the mechanisms of action of acupuncture in this type of injury and to prove that the results obtain are fully related with the treatments applied.

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## **Chapter 5.**

### **Research in acupuncture's short-term effects & Future Perspectives**



The present thesis have pointed out that the effects of acupuncture may happen in a short-term time-frame and even shorten the time needed to achieve a certain health status when compared with the actual western knowledge. Although it is evident that acupuncture “sends a signal” and a physiological response is given (for example, a change in electric conductivity of the skin – see chapter 3), the mechanism of action is not yet fully understood under the current human physiology knowledge. Additional research in this field is needed.

The next step of investigation, in the line of research of the present thesis, should address the physiological pathways that acupuncture uses to induce any type of effect in the human body. Several core questions should be addressed in future research, as follow:

- Does local and distal acupuncture use the same or different physiological pathways?
- Have different acupoints specific reflex of different regions in the body?
- Are the acupuncture response time inhibited or improved by drugs or natural extracts?
- Does acupuncture use more than a pathway (e.g. nervous system, endocrine systems) to obtain effects in different time frames?
- How much time an acupuncture treatment may last? Can it be permanent?

Additionally to this research process, an effort to convert empirical language of TCM to a scientific way of describing the physiology and physiopathology would be great improve in delivering knowledge to future students in life science and health related courses.

We understand that limitations to continue the research will include the access to advanced diagnosis systems, sample, and the ability to produce double blinded trials. However, as ICBAS-UP is the sole institution in Portugal that produce research in TCM, it is expected that the TCM’s cutting edge research in Portugal should be produced at ICBAS-UP.

I personally hope that the present thesis may impel other students and researchers to gather new ideas and continue to produce research in TCM, and specifically in acupuncture. There is a growing need to have qualified people in both clinical and research context to elevate the quality of TCM research and teaching in Portugal and specially at ICBAS-UP turning this institution in the main Portuguese reference in this field.





## Supporting Information



## Appendix I – Details of normalized frequencies for SMD of both control and experimental group

The Table S1 is supporting information for the Figure 5 in the main manuscript. It presents the normalized frequency values of SMD for both control and experimental groups.

**Table S1 – Control and experimental group results:** Normalized frequency values of the control and experimental groups, concerning the split-meridian differences ( $0 < \text{SMD} < 35+$ ) for the three measurement moments of the experiment (M1 – phase 2, M2 – phase 4 and M3 – phase 5).

	Measurement	SMD ( $\mu\text{A}$ )							
		0	5	10	15	20	25	30	35+
Experimental	M1	0,00	0,00	0,00	0,00	0,45	0,30	0,15	0,10
	M2	0,30	0,20	0,25	0,15	0,00	0,00	0,05	0,05
	M3	0,45	0,40	0,10	0,05	0,00	0,00	0,00	0,00
Control	M1	0,00	0,00	0,00	0,00	0,33	0,17	0,42	0,08
	M2	0,00	0,08	0,00	0,17	0,25	0,00	0,25	0,25
	M3	0,00	0,08	0,00	0,00	0,33	0,08	0,17	0,33

